vacuum lines do not become contaminated.

(8) Test tubes. (i) Tubes containing viable etiologic agents should be manipulated with extreme care. Studies have shown that simple procedures, such as removing a tube cap or transferring an inoculum, can create a potentially hazardous aerosol.

(ii) Manipulation of biohazardous test tubes will be conducted in biological safety cabinets. Tubes and racks of tubes containing biohazardous material should be clearly marked. The individual employee must ensure that tubes containing biohazardous material are properly sterilized prior to disposal or glassware washing. Safety test tube trays should be used in place of conventional test tube racks to minimize spillage from broken tubes. When safety test tube trays are not used, the conventional test tube racks will be placed in a tray large enough to contain any potential spill. A safety test tube tray is one having a solid bottom and sides deep enough to hold all liquids, should a test tube break.

(9) Care should be exercised when using membrane filters to obtain sterile filtrates of viable etiologic agents. Due to the fragility of the membranes and other factors, such filtrates cannot be considered noninfectious until laboratory culture or other tests have proven their sterility.

(10) The preparation, handling, and use of dry powders of viable etiologic agents in open containers presents unusual hazards. The slightest manipulation of such powders can cause the generation of aerosols containing a high concentration of etiologic agents. Therefore, work with dry powders of etiologic agents in open containers should be carried out in gas-tight biological safety cabinets.

§ 627.13 Biosafety level 1.

(a) *Requirements beyond those for all etiologic agents.* BL–1 operations follow the general techniques described in §§ 627.12(a) and 617.12(b).

(b) *Additional laboratory requirement.* Contaminated materials that are to be decontaminated at a site away from the laboratory are placed in a durable leak-proof container which is closed before being removed from the laboratory. Examples of suitable containers are metal tubs with lids or plastic bags that are sealed and then placed inside a rigid container for transport.

(c) *Additional animal requirements.* (1) Bedding materials from animal cages will be removed in such a manner as to minimize the creation of aerosols and disposed of in compliance with applicable institutional or local requirements.

(2) Cages are washed manually or in a cagewasher. Temperature of final rinse water will be a minimum of 180 °F.

(3) Laboratory coats, gowns, or uniforms worn in animal rooms shall not be worn in other areas.

§ 627.14 Biosafety level 2.

(a) Additional requirements. In addition to the general microbiological techniques stated in § 627.13, BL–2 operations include the following requirements:

(1) When etiologic agents are in use, a hazard warning sign incorporating the universal biohazard symbol is posted on the access door of the work area. The hazard warning sign identifies the etiologic agent, lists the name and telephone number of the institute director or other responsible person(s), and indicates the special requirement(s) for entering the laboratory.

(2) Animals not involved in the work being performed are not permitted in the laboratory.

(3) Special care is taken to avoid skin contamination with the etiologic agents; gloves will be worn when handling etiologic agents or infected animals.

(4) All wastes from laboratories and animal rooms are decontaminated before disposal.

(5) Hypodermic needles and syringes are used only for parenteral injection and aspiration of fluids from laboratory animals and diaphragm bottles.

(6) Spills and accidents which result in a potential exposure to etiologic agents will be reported immediately to the safety officer, the project leader, and the institute director.

(7) Biological safety cabinets (Class I or II) will be used when:

(i) Procedures with a high potential for creating infectious aerosols are conducted.

(ii) High concentrations or large volumes of etiologic agents are used.

(8) Laboratory coats, gowns, smocks, or uniforms will be removed before leaving the animal facility or laboratory area.

(b) Additional animal requirements.

(1) Cages must be decontaminated, preferably by autoclaving, before they are cleaned and washed.

(2) Approved molded masks are worn by all personnel entering animal rooms housing nonhuman primates.

(3) If floor drains are provided, the drain traps will be kept filled with water or a suitable disinfectant.

§ 627.15 Biosafety level 3.

(a) *Additional requirements.* In addition to the requirements stated in §§ 627.13 and 627.14, the following requirements apply—

(1) Approved molded masks or respirators with HEPA filters are worn by all personnel in rooms housing infected animals.

(2) Protective clothing worn in a laboratory or animal room will be removed before exiting the laboratory or animal room.

(3) Clothing worn in laboratories and animal areas to protect street clothing will be decontaminated before being laundered.

(b) *Additional laboratory requirements.* (1) Laboratory doors will be kept closed.

(2) All activities involving etiologic agents will be conducted in biological safety cabinets (Class I, II, or III) or other physical containment devices within the containment module. No work in open vessels is conducted outside a biological safety cabinet.

(3) The work surfaces of biological safety cabinets and other containment equipment will be decontaminated after work with etiologic agents. Plastic-backed paper toweling should be used on nonperforated work surfaces within biological safety cabinets to facilitate clean-up.

(c) *Additional animal requirements.* (1) Cages are autoclaved before bedding is removed and before they are cleaned and washed.

(2) Gloves are removed aseptically and autoclaved with other wastes before being disposed of or reused.

(3) Boots, shoe covers, or other protective footwear and disinfectant foot baths must be available and used when indicated.

(4) Personal protective clothing and equipment and other physical containment devices are used for all procedures and manipulations of etiologic agents or infected animals. The risk of infectious aerosols from infected animals or their bedding shall be reduced by housing animals in partial containment caging systems as described in § 627.56.

(d) *Work with BL–3 etiologic agents that require additional secondary containment.* Facilities in which work with certain viruses, for example, Rift Valley fever, yellow fever, and Venezuelan equine encephalitis, is conducted require HEPA filtration of Xallexhaust air prior to discharge from the laboratory. All persons working with those agents for which a vaccine is available should be immunized.

§ 627.16 Biosafety level 4.

Laboratory work at BL–4 must follow the requirements stated in §§ 627.13, 627.14 and 627.15 as well as the following:

(a) All activities are conducted in Class III biological safety cabinets or in Class I or II biological safety cabinets in conjunction with a one-piece positive pressure personnel suit ventilated by a life-support system.

(b) Biological materials to be removed from the Class III cabinet or from the maximum containment laboratory in a viable or intact state must be transferred to a sealed nonbreakable primary container, enclosed in a nonbreakable sealed secondary container, and removed from the facility through a disinfectant dunk tank, fumigation chamber, or an airlock designed for this purpose.

(c) No materials, except for biological materials that are to remain in a viable or intact state, are removed from the maximum containment laboratory unless they have been autoclaved or decontaminated before they leave the facility. Equipment or material which might be damaged by